JOURNAL OF NATURAL REMEDIES
DOI: 10.18311/jnr/2025/44457
RESEARCH ARTICLE

Evaluation of Synergistic Antifungal Activity of *Curcuma longa* (Rhizomes) and *Mangifera indica* (Leaves)

Praveena Gandhi*, Gayathri Kumararaja, K. Muhammed Jaseel, Muhammed Hamras, K. Mohamed Basith, A. Mohammed Asarudeen, S. Meena and R. Marimuthu

Faculty of Pharmacy, Karpagam Academy of Higher Education, Coimbatore - 641021, Tamil Nadu, India; pravina.gj@gmail.com

Abstract

Background: The ongoing study proceeded to investigate antifungal properties for a combined extract of leaves of *Mangifera indica* L. and rhizomes of *Curcuma longa* L. *M. indica* Linn both had a salient role in the Indian traditional system. The leaves of *M. indica* L were reported to have good medicinal values in the *Ayurveda, Siddha* and *Unani* systems of medicine. Mango leaf posse sses various effects like antifungal, antidiabetic, analgesic, antibacterial and anti-inflammatory properties. Turmeric (*C. longa* L.) is greatly used as a dominant cosmetic and aromatic purpose throughout the India, This was habitually used by many since from olden days in *Ayurvedic* compositions, this miraculous drug has been widely used for various diseases like skin issues, anticancer, CVS, respiratory-related problems, antifungal and cough. **Aim:** To study the synergistic effect of *M. indica* and *C. longa* for their antifungal properties. **Methods:** Hydroalcoholic extraction was done to evaluate the antifungal activity of the combined products. The extraction of products was done by using soxhlet extraction. Zone of inhibition was done to investigate the medicinal values of extracts. *Candida albicans* species were used to investigate the antifungal activity of the extracts. **Results:** The *M. indica*: *C. longa* (1:1) combination showed the highest antifungal activity (25±1.3 mm) against *C. albicans*, surpassing individual extracts (16±3.8 mm, 19±5.2 mm) and other ratios. Fluconazole exhibited 30±4.6 mm inhibition. This highlights the enhanced antifungal potential of the combined extracts. **Conclusion:** The investigation have shown the extensive antifungal activity of *C. longa* and *M. indica*, with the support of *in vitro* studies it is revealed that the combined extracts of *M. indica* and *C. longa* shows synergistic antifungal activity.

Major Findings: This study highlights the enhanced antifungal potential of combining *Mangifera indica* and *Curcuma longa* (1:1), which showed a significant inhibition zone (25±1.3 mm) against *Candida albicans*. The synergy between these extracts outperformed individual effects, demonstrating their promising therapeutic value as a natural antifungal alternative.

Keywords: Antifungal Activity, Combined Extract, *C. longa, M. indica*, Zone of Inhibition

1. Introduction

A fungus is a multi-cellular creature. Fungi include yeast, molds, and mushrooms. Most fungus can be seen with the naked eye. However, some are microscopic organisms. Some microscopic fungi, such as yeast, are utilized in the food and beverage sector to make bread, beer, and wine. Other kinds of fungi serve an important part in the pharmaceutical and biotechnology sectors by generating antibiotics and enzymes. Some fungi are used as probiotics[1]. Some fungi are harmful to animals (including humans) and plants. *C. albicans*, *A. fumigatus*, and *C. neoformans* are medically important pathogenic fungus. An antifungal agent is a medication that selectively eliminates fungal infections from a host while being minimally hazardous to the host. These can be herbal or synthetic. There have been reports of growing medicine resistance in human illnesses, as well as adverse consequences from certain antimicrobial treatments. Failure to treat, especially with the present

*Author for correspondence

Article Received on: 12.06.2024 Revised on: 19.02.2025 Accepted on: 19.02.2025

growing trends of Multi-Drug Resistance (MDR) to accessible modern medications or antibiotics among emerging and re-emerging bacterial infections, poses substantial hazards. As a result, it is critical to seek out novel agents that are better, cheaper, and have no side effects while treating infectious diseases, particularly in developing countries[2,3]. Plants are the cheapest and safer alternative sources of antimicrobials, so herbal medicines as the principal remedy in ancient system of medicine have been applied in medical practices from antiquity and in the contemporary scenario herbal medicines are the most emerging field in the pharmaceutical industry[4]. We are interested in the antifungal synergy of turmeric extract and mango leaf extract. Turmeric derived from *C. longa* L. (*Zingiberaceae*) has previously been found to suppress microorganisms. *C. longa* L. (turmeric) has long been used as both a home medication and a spice in Southeast Asia[2]. Mango leaf extract is prepared from *M. indica* L. (*Anacardiaceae*), which has previously been shown to suppress microorganisms. Traditionally, the mango plant and its many parts, such as bark, fruit, and leaf, have therapeutic properties[3]. Although these plants are well-documented medicinal species, variations in phytochemical composition due to environmental and geographical factors necessitate further confirmation. Pharmacognostic and phytochemical evaluations ensure standardization, validate bioactive compounds, and enhance the reproducibility of antifungal efficacy. Thus, the current study was carried out to investigate the synergistic antifungal action of turmeric and mango leaf inhibits *Candida* spp.

2. Materials and Methods

2.1 Pharmacognostic Studies

Standardization strives to provide consistent potency and control over the full range of bioactive chemical components found naturally in medicinal plants from batch to batch. Using physical properties to authenticate raw medications can aid in identifying botanical species. The tests were conducted to determine the physicochemical criteria for total ash, foreign organic matter, and moisture content[5,6]. Authentication and standardization are required procedures in traditional medical systems, particularly for herbal medications and their formulations[7]. Evaluation of a drug entails confirming its identity, determining its purity, and detecting the kind of adulteration. Depending on the morphological and microscopical examinations of crude pharmaceuticals, quality control can be attempted using various evaluation methods such as physical, chemical, and biological behaviours. Drug description includes the systematic identification of crude pharmaceuticals as well as quality assurance. Pharmacognostical investigations involve identification, authentication, and standardization using morphological, microscopical, and physicochemical observations in accordance with WHO criteria. Standardization of herbal and herbal medications begins with proper sample identification, organoleptic evaluation, pharmacological evaluation, volatile matter estimation, quantitative evaluation, phytochemical evaluation, toxicity testing, and biological activity.

2.1.1 Macroscopy

The shade-dried leaves and rhizomes were finely pulverized before being passed through filter no.60 for microscopic inspection. Organoleptic characteristics were observed. To determine the numerous distinguishing traits, various staining reagents were used. Powdered samples were washed with NaOH, mounted in glycerin medium, dyed with 1% phloroglucinol in 90% ethanol, concentrated hydrochloric acid, and viewed under a microscope. Phloroglucinol was used to dye all of the lignified cells pink. The plant material is classified depending on its sensory properties. Organoleptic and morphological examination is the easiest and quickest method for determining a samples identification, purity, and quality. As a result, this observation is crucial before moving forward with any more testing.

2.1.2 Microscopy

Fresh *C. longa* rhizomes were analyzed under a microscope. The parts were hand cut using free sectioning. Several mounts of the specimens microscopical segment were made and inspected under a microscope. Similarly, fresh *M. indica* leaves were exposed to microscopy. The parts were cut using free sectioning. Numerous mounts of the specimens microscopical section were prepared and examined under a microscope[8].

2.1.3 Powder Microscopy

The shade-dried leaves and rhizomes were finely pulverized before being passed through filter no.60 for microscopic inspection. Organoleptic properties were observed. To determine the numerous distinguishing traits, various staining reagents were used. Powdered components were rinsed with NaOH, placed into glycerin medium, stained with 1% phloroglucinol in 90% ethanol, concentrated hydrochloric acid, and examined under a microscope. Phloroglucinol was used to dye all of the lignified cells pink[9].

2.2 Phytochemical Studies

Non-nutritive molecules found in plants called phytochemicals include substances that protect and avoid diseases. They are involved in a variety of processes, among which are those that help prevent cell damage, cancer cell multiplication, and antifungal activity. Certain phytochemicals work as anti-oxidants, whereas another inhibit enzymes. These substances are evaluation is carried out in order to prove the phytoconstituents found in plants employing an appropriate quantitative assay. The plants medicinal properties must be thoroughly investigated. The current phytochemical study of *M. indica* leaves and *C. longa* rhizomes includes a qualitative component.

2.2.1 Preparation of Methanolic Extracts

Extraction is the first stage in phytochemical investigations. It draws out the metabolites into the extraction solvent based on its polarity. The initial step was to prepare successive solvent extracts. The dried coarsely powdered samples of *C. longa* and *M. indica* were extracted separately with hydroalcoholic solvent using methanol (60-80 °C) in a Soxhlet system. Each extract was concentrated using evaporation. The percentage yield, colour, and consistency of these extracts were recorded and followed by more comprehensive phytochemical and *in-vitro* antifungal activities[10].

2.2.2 Phytochemical Screening

2.2.2.1 Qualitative Analysis of Phytochemical Constituents

To identify the existence of different botanical compounds parts, both the extracts and dehydrated powders received qualitatively phytochemical testing[11,12].

2.2.2.2 Antimicrobial Activity

Candida albicans is maintained as a fungal culture on Sabouraud Dextrose Agar (SDA). *In vitro* activity was measured using the well diffusion method. SDA was put into sterilized Petri plates and let to solidify. Wells with a diameter of 9mm were formed on the plates, and the extracts were applied to distinct wells in serially increasing amounts of 100µg/ml, 75µg/ml, and 50µg/ml for individual extracts, and combined extracts in the ratio of 1:1, 3:1, 1:3. (*M. indica*: *C. longa*) by using a micropipette. Fluconazole (10µg) was employed as a reference, while solvents served as negative controls. The plates were labelled, covered, and incubated at 28°C for 24 hours. The extracts activity was assessed by measuring the diameter of the zone of inhibition[11].

3. Results and Discussion

This study found antifungal efficacy in a combination extract of *M. indica* and *C. longa*. The test organisms were resistant to antifungal action from the mixed extracts. *C. albicans* organisms were sensitive to treatment with the common antifungal medication fluconazole at all concentrations tested are presented in Table 3. The qualitative phytochemical analysis of both *M. indica* and *C. longa* extracts, summarized in Table 1 and Table 2, identified key bioactive compounds, such as flavonoids, alkaloids, and terpenoids. These compounds are likely contributing to the antifungal properties observed in the extracts.

3.1 Pharmacognostical Studies

The Pharmacognostic study results of *M. indica* are as follows;

3.1.1 Organoleptic Characters

Nature : Fine coarse powder
Colour : Shiny and dark green
Odour : Characteristic
Taste : Bitter

3.1.2 Morphological Characters

Type : Alternate, simple
Shape : Oblong or Elliptic-lanceolate
Size : 15-35 cm long 6-16 cm broad as shown in Figure 1.

Surface : Adaxial and Abaxial as illustrated in Figure 2.
Margin : Entire
Apex : Cuneate-subacute
Base: Acuminate

3.1.3 Anatomy of Leaf

An epidermal layer is present on both the upper and lower surfaces. The upper and lower epidermal layers are one celled thick and made up of barrel-shaped, as displayed in Figure 3, compactly packed cells. Epidermal cells have a thick cuticle on their exterior surfaces. In comparison, the top epidermis has a thicker cuticle. The stomata are only found on the lower epidermis.

3.1.4 Powder Microscopy

The powder microscopy of *M. indica* Linn., showed the following characteristics.



Figure 1. Length and width of the leaf of *M. indica.*



Figure 2. Adaxial and abaxial surface of *M. indica.*

3.1.5 Organoleptic Characters

Nature : Fine coarse powder
Colour : Shiny and dark green colour
Odour : Characteristic odour
Taste : Astringent taste

3.1.6 Powder Microscopic Characters

The powder of the leaves shows the following inclusions, when examined under the microscope covering trichomes in Figure 4 and lignified cells in Figure 5.

3.1.7 Pharmacognostical Studies C. longa

The results of pharmacognostic studies are as follows of *C. longa*

3.1.8 Organoleptic Characters

Nature : Fine coarse powder
Colour : Deep orange-yellow inside as displayed in Figure 6
Odour : Aromatic
Taste : Aromatic and bitter

3.1.9 Morphological Characters

Colour : Deep orange-yellow inside
Length : Medium sized, 2-3 X 2-3 cm as shown in Figure 7
Shapen : Conical
Surface : Root scars and annulations are seen.
Fracture : Horny
Odour : Aromatic
Taste : Aromatic and bitter

3.1.9 Anatomy of Rhizomes

The rhizome is tuberous, possesses a camphoraceous aromatic smell, and measures approximately 2-6 cm

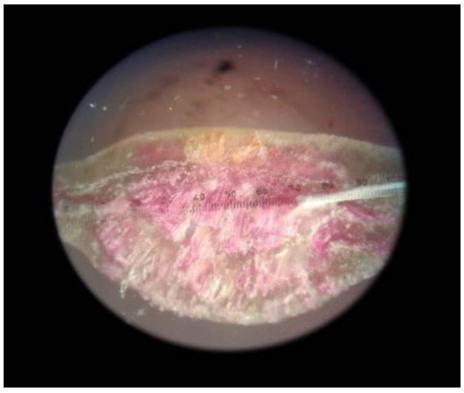


Figure 3. Transverse section of the leaf of *M. indica.*

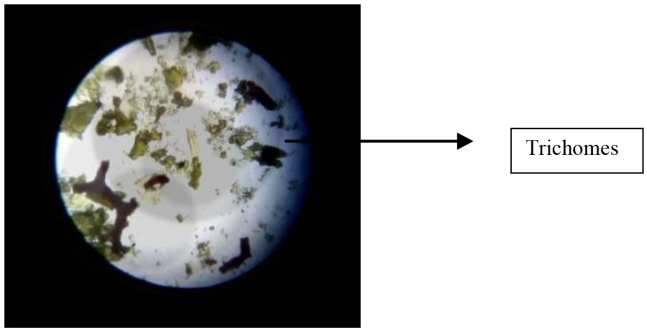


Figure 4. Unicellular covering trichomes under microscope.

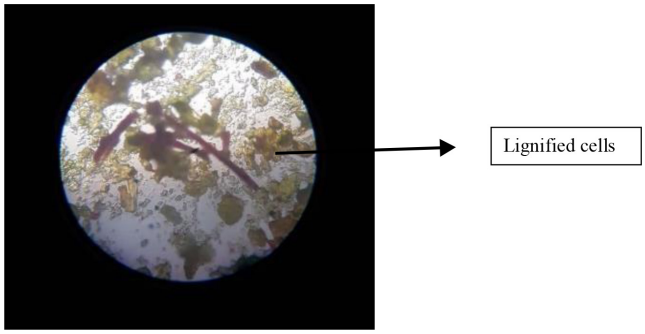


Figure 5. Pink coloured lignified cells under microscope.



Figure 6. Rhizomes of *C. longa*.

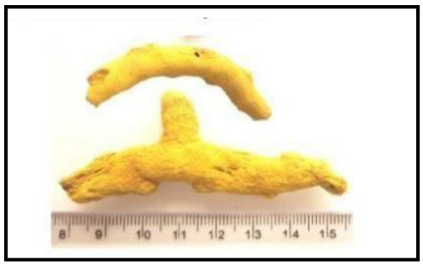


Figure 7. Length of the rhizome.

in diameter. The size and form of it seem to change. It is a sessile horizontally flattened out, and covered with intrusive roots, root scars, and warts; as shown in Figure 6, it also includes transverse circular creases on its outer layer that resemble node and internodal regions on the rhizome. The rhizomes surface (cork) is dark brown, bluish black, or buff in color, with circular collections of crusty leaf leftovers which simulate development circles. The branching arrangement is more or less symmetrical which is illustrated in Figure 8.

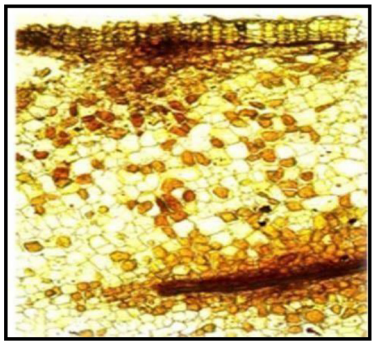


Figure 8. Anatomy of the rhizome of *C. longa*.

3.1.10 Powder Microscopy

The powder microscopy of *C. longa* showed the following characters.

Organoleptic Characters

Nature	: Fine coarse powder
Colour	: Deep orange-yellow inside
Odour	: Aromatic
Taste	: Aromatic and bitter

3.1.11 Microscopical Characters

The powder of the rhizomes shows the following inclusions when examined under the microscope

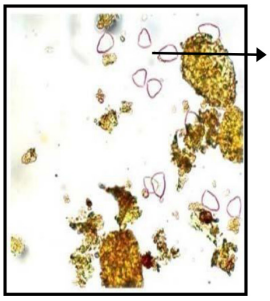


Figure 9. Pitted tracheids view of *C. longa* under the microscope.

presence of pitted tracheids, starch grains, and fibres are displayed in Figures 9, 10, 11 respectively.

3.2 Qualitative Phytochemical Analysis of *M. indica*

Table 1. Preliminary phytochemical examination of *M. indica* extracts

Si.No.	Chemical Constituents	Methanol Extract
1	Alkaloids	+
2	Glycosides	+
3	Steroids	+
4	Flavonoids	-
5	Carbohydrates	-
6	Glycosides	+
7	Saponins	+
8	Triterpenoids	+
9	Protein and amino acids	-
Note: (+) indicates presence, (-) indicates absence.		

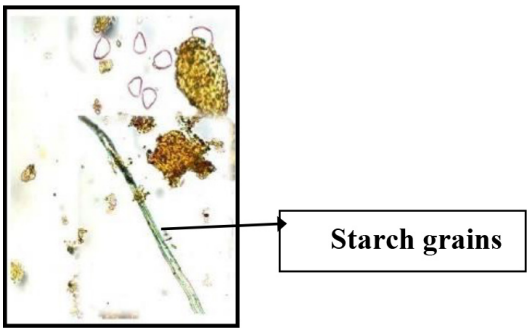


Figure 10. Starch grains view of *C. longa* under the microscope.

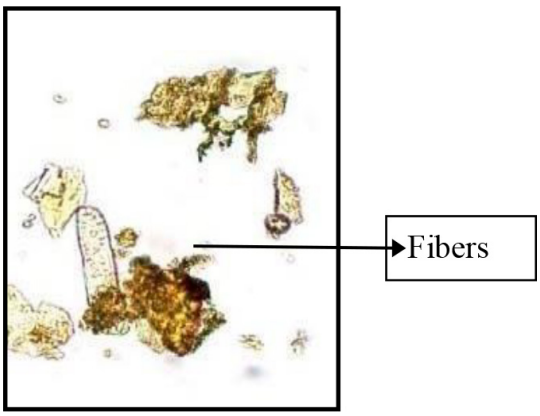


Figure 11. Fibers view of *C. longa* under the microscope.

3.2.1 Qualitative Phytochemical Analysis of *C. longa*

Table 2. Preliminary phytochemical examination for extracts of *C. longa*

Si.No.	Chemical Constituents	Methanol Extract
1	Alkaloids	+
2	Flavonoids	+
3	Triterpenoids	+
4	Protein and amino acids	+
5	Glycosides	+
6	Saponins	+
7	Phenolic compounds and Tannins	+
8	Carbohydrates	+
9	Steroids	+
Note: (+) indicates presence, (-) indicates absence		

3.3 *In Vitro* Antifungal Activity

Table 3. The antifungal activities (diameter of inhibition zone, mm) of the methanol extracts of *M. indica*, *C. longa* and their combined extracts against *C. albicans*

Tested Exract	Diameter of Inhibition Zone (mm)
M. indica extract (100 µg/ml).	16±3.8
M. indica extract (75 µg/ml).	10±4.7
M. indica extract (50 µg/ml).	6±3.1
C. longa extract (100 µg/ml)	19±5.2
C. longa extract (75 µg/ml)	15±1.5
C. longa extract (50 µg/ml)	8±3.46
Combined Extracts (1 *M.vindica*: 1 *C. longa*)	25±1.3
Combined Extracts (3 *M. indica*: 1 *C. longa*)	9±5.6
Combined Extracts (1 *M. indica*: 3 *C. longa*)	13±3.4
Fluconazole (Reference)	30±4.6

S=Standard drug; C1=Concentration100µg/ml; C2=CONCentration75µg/ml;

C3 = Concentration 50µg/ml

Three different concentrations of *M. indica* extracts show significant antifungal activity against



Figure 12. Zone of inhibition of different concentrations of *M. indica.*



Figure 13. Zone of inhibition of different concentrations of *C. longa.*

C. albicans, 100µg/ml showed better antifungal activity as illustrated in Figure 12, compared to 75 µg/ml and 50 µg/ml with standard drug fluconazole of 10µg/ml.

S = Standard drug;

C1 = Concentration 100µg/ml;

C2 = Concentration 75µg/ml;C3 = Concentration 50µg/ml

Three different concentration of *C. longa* extracts shows significant antifungal activity against *C. albicans*, 100µg/ml showed better antifungal activity as shown in Figure 13, compared to 75 µg/ml and 50 µg/ml with standard drug fluconazole of 10µg/ml..

S = Standard drug**;**

C1 = Concentration 1 (*M. indica***:** *C. longa,1:1***);**

C2 = Concentration 2 (*M. indica***:** *C. longa,3:1***);** C3 = Concentration 3 (*M. indica*: *C. longa,1:3*)

Synergistic antifungal activity is obtained for combined extracts. The combined extract *M. indica*:*C. longa* 1:1 showed maximum synergistic antifungal activity when compared to combined extracts in theratio *M. indica*:



Figure 14. Zone of inhibition of different ratios of combined extracts of *M. indica* and *C. longa.*

C. longa, 3:1 and *M. indica*: *C. longa,* 1:3 and individual extracts of *M. indica* and *C. longa* with standard drug fluconazole of 10µg/ml as observed in Figure 14.

4. Conclusion

Pharmacognostical evaluation including macroscopy, microscopy, and powdered microscopy, as well as phytochemical evaluation, have been examined to gather data on the features of the plants *M. indica* and *C. longa*. In the laboratory investigations on Candida albicans show that individual extracts have antifungal action against Candida albicans at high concentration. Several previous studies have shown the substantial antifungal activity of *C. longa* extract and *M. indica*; nevertheless, *in vitro* experiments have demonstrated that the combined extracts of *M. indica* and *C. longa* exhibit synergistically antifungal activity. More study is needed to determine the active ingredients in both botanical extracts and the formulations they contain so that they can be utilized as other substances in the treatment of harmful bacteria and fungus. The findings may be important, demonstrating the curative properties of both plants utilized in conventional medicine. It could serve as an appropriate foundation on subsequent botanical compounds and pharmacological investigations.

5. Author Contribution

The authors collectively contributed to the study's design, experimentation, data analysis, and interpretation. Manuscript preparation and revisions were carried out collaboratively. All authors reviewed and approved the final version of the work.

6. References

1. Zanello G, Meurens F, Berri M, Salmon H. *Saccharomyces boulardii* effects on gastrointestinal diseases. Curr Issues Mol Biol. 2009; 11:47-58. https://doi.org/10.21775/cimb.011.047
2. Banerjee A, Nigam SS. Antimicrobial efficacy of the essential oil of *Curcuma longa*. Indian J Med Res. 1978; 68:864-866.
3. Hamuel J, Doughari J, Obidah JS. Antibacterial potentials of stem bark extracts of *Leptadenia lancifolia* against some pathogenic bacteria. PhOL. 2008; 3:172-180.
4. Olasehinde GI, Sholotan KJ, Openibo JO, Taiwo OS, Bello OA, Ajayi JB, *et al*. Phytochemical and antimicrobial properties of *M. indica* leaf extracts. CJPLS. 2018; 6(1).
5. Divya A. Pharmacognosy: Facts and future. Res Rev J Pharmacogn Phytochem. 2015; 2(3).
6. Amphawan A, Vanittanakom N, Buddhasukh D. Antifungal activity of turmeric oil extracted from *C. longa* (*Zingiberaceae*). J Ethnopharmacol. 1995; 49(3):163-165. https://doi.org/10.1016/0378-8741(95)01320-2
7. Shahin N, Alam S, Ali M. Pharmacognostical standardisation and antidiabetic activity of *Artocarpus heterophyllus* leaves Lam. Int J Drug Dev Res. 2012; 4(1):60-65.
8. Nagani KV, Kevalia J, Chanda SV. Pharmacognostical and phytochemical evaluation of stem of *Cissus quadrangularis* L. Int J Pharm Sci Res. 2011; 2(11):2872-2876.
9. Duraisankar M, Ravindran AD. Identification of *Curcuma longa* rhizomes by physicochemical and TLC fingerprint analysis. Int J Pharm Tech Res. 2015; 8(6):913-919.
10. Kokate CK. Practical pharmacognosy. 5th Edition. Vallabh Prakashan. 2014.
11. Harborne JB. Phytochemical methods: A guide to modern techniques of plant analysis. 2nd Edition. Chapman and Hall. 1973.
12. Pamila UA, Karpagam S. Preliminary phytochemical screening and antimicrobial activity of cotyledon of two varieties of *Mangifera indica* L. Int J Sci Res. 2018; 7(5):122-125.